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## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

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<b>(21) International Application Number:</b> PCT/IB00/00393  <b>(22) International Filing Date:</b> 21 March 2000 (21.03.00)  <b>(30) Priority Data:</b> 9906715.9      23 March 1999 (23.03.99)      GB  <b>(71) Applicant (for all designated States except US):</b> FERRING B.V. [NL/NL]; Polaris Avenue 144, NL-2132 JX Hoofddorp (NL).  <b>(72) Inventor; and</b> <b>(75) Inventor/Applicant (for US only):</b> BROQUA, Pierre [FR/FR]; 18, rue des Bergeronnettes, F-01710 Thoiry (FR).  <b>(74) Agent:</b> GEERING, Keith, Edwin; Reddie & Grose, 16 Theobalds Road, London WC1X 8PL (GB).		<b>(81) Designated States:</b> AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).  <b>Published</b> <i>Without international search report and to be republished upon receipt of that report.</i>
<b>(54) Title:</b> COMPOSITIONS FOR PROMOTING GROWTH  <b>(57) Abstract</b>  Inhibitors of dipeptidyl peptidase IV and pharmaceutical compositions comprising these inhibitors are useful in the treatment of short stature due to Growth Hormone deficiency and for promoting GH-dependent tissue growth or regrowth.		

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### Compositions for Promoting Growth

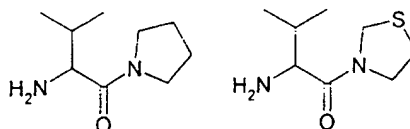
The present invention relates to agents and compositions for promoting animal growth, e.g. in underdeveloped humans, and for accelerating tissue repair and regeneration.

#### *Inhibitors of DP-IV*

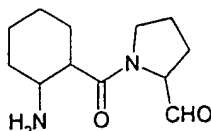
Dipeptidyl peptidase IV (DP-IV, also dipeptidyl aminopeptidase IV, DPP-IV, DAP-IV, EC 3.4.14.5) is a serine peptidase that cleaves the amino-terminal dipeptide from peptides and proteins. It recognises substrates wherein the N-terminal sequence is X-Pro or X-Ala. Inhibitors of DP-IV have been proposed as therapeutic agents for the treatment of inflammatory diseases and AIDS. Generally, the known inhibitors of DP-IV are analogues of the substrate. Examples of DP-IV inhibitors are those disclosed in DD 296 075 A5 (Neubert *et al.*, November 1991), WO91/16339 (Bachovchin *et al.*, October 1991), WO93/08259 (Bachovchin *et al.*, April 1993), WO95/15309 (Jenkins *et al.*, June 1995), WO98/19998 (Villhauer, May 1998), WO99/46272 (Scharpe *et al.*, September 1999) and WO99/61431 (Demuth *et al.*, December 1999). Prodrugs of some of these inhibitors have also been described in WO99/67278 and WO99/67279 (both Demuth *et al.*, December 1999).

The following table sets out general types of DP-IV inhibitor compounds, and specific examples thereof which are amongst those preferred for use in the present invention; it also indicates the patent publications from whose broader range of disclosed compounds these types and examples are drawn. It is emphasised that all DP-IV inhibitors disclosed in the quoted DD and WO specifications can be used in the present invention, and reference is positively directed to these prior specifications for full information on the general and more specific formulae and individual compounds concerned. For example, in the table below the indicated pyrrolidine and thiazolidine rings can be replaced by a wide range of other heterocycles of various ring sizes and/or the indicated aminoacyl moieties can be replaced by a wide range of others, as taught by the indicated publications, to give other DP-IV inhibitors for use in the present invention.

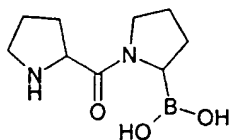
Amino-acyl pyrrolidides and thiazolidides (see DD 296 075 A5), e.g.



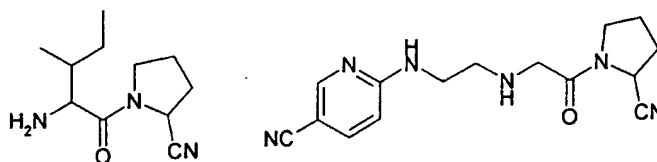
Amino-acyl pyrrolidine aldehydes (see DD 296 075 A5 and WO95/15309), e.g.



Amino-acyl pyrrolidine boronic acids (see WO91/16339 and WO93/08259), e.g.



Amino-acyl pyrrolidine nitriles (see WO95/15309 and WO98/19998), e.g.



#### *Actions of Growth Hormone*

Growth Hormone (GH, Somatotropin) is secreted by the pituitary in response to a hypothalamic signal in the form of Growth Hormone Releasing Hormone (GHRH). The most abundant isoform of human GH is a 191-residue peptide and GHRH is a 44-residue peptide. GH is a key factor in promoting the normal development of children, and GH deficiency can lead to dwarfism. In adults the importance of GH is less well defined, although plasma levels of GH are similar in adults and children. It is possible that GH plays a role in tissue repair following injury.

The current therapeutic options for GH deficiency include the administration of synthetic GH or of GHRH. However, since both these hormones are peptidic, it is not possible to give them orally. Administration is usually by injection, which can be distressing for the patient (and particularly for children) despite the development of needleless injection systems.

Furthermore, a single injection of GHRH is only an approximation of the pulsatile release of this factor from the hypothalamus. Finally, because both are relatively large peptides, both GHRH and particularly GH are expensive to produce. In consequence, there exists an unmet need for a therapeutic agent that will promote the actions of GH. We have now found that DP-IV inhibitors can increase the concentration of circulating GH.

A first aspect of the present invention is a pharmaceutical composition for the treatment of dwarfism and sub-normal growth (e.g. in children), and for promoting tissue repair (e.g. following injury), which composition is characterized by the inclusion of inhibitor of DP-IV. A second aspect of the present invention is a novel use of inhibitors of DP-IV, namely in the treatment of dwarfism and tissue injury. A third aspect of the invention is an improved protocol for accelerating the growth of children and the regeneration of injured tissue, wherein the subject is administered a composition comprising DP-IV inhibitor.

The use of DP-IV inhibitors in this way presents many advantages over current treatment regimens that involve GH or GHRH. GHRH and particularly GH are large peptides that are either isolated from natural sources (such as animal brain preparations) or prepared in culture using recombinant cells. Isolation from animal sources requires that attention be paid to risk of disease transmission and the presence of antigenic protein contaminants. Recombinant hormones are less likely to transmit human pathogens but are still potentially contaminated with antigenic protein. GHRH can be prepared synthetically, but such a synthesis is expensive and necessitates careful purification of the product. In contrast, DP-IV inhibitors are small molecules that are readily accessible using standard synthetic methods. They are non-antigenic, easy to purify and inexpensive.

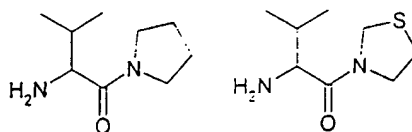
A further advantage is that DP-IV inhibitors are in many cases biologically active after oral administration. This is in contrast to GH and GHRH, which must be administered by injection. Hence the use of DP-IV inhibitors leads to a less invasive protocol that is less stressful for the patient.

In a first aspect, the present invention provides a pharmaceutical composition for the treatment of sub-normal development or dwarfism. The composition is particularly effective for the treatment of sub-normal development or dwarfism due to growth hormone deficiency. The invention equally provides a composition for the promotion of tissue regeneration.

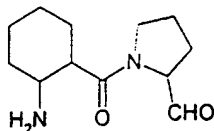
These compositions are characterized in that they comprise inhibitor of DP-IV. The compositions may further include such pharmaceutically acceptable excipients as are generally known in the art, such as diluents, carriers, bulking agents, binding agents, dispersants, stabilizers and the like.

In the context of the present invention, a compound is considered to be an inhibitor of DP-IV if it inhibits the action of the enzyme at a concentration of  $1\mu\text{M}$ . Preferably, such a compound inhibits the action of DP-IV at concentrations below  $100\text{nM}$  and does not inhibit other enzymes at concentrations below  $1\mu\text{M}$ . The following table sets out general types of DP-IV inhibitor compounds, and specific examples thereof which are amongst those preferred for use in the present invention; it also indicates the patent publications from whose broader range of disclosed compounds these types and examples are drawn. It is emphasised that all DP-IV inhibitors disclosed in the quoted DD and WO specifications can be used in the present invention, and reference is positively directed to these prior specifications for full information on the general and more specific formulae and individual compounds concerned. For example, in the table below the indicated pyrrolidine and thiazolidine rings can be replaced by a wide range of other heterocycles of various ring sizes and/or the indicated aminoacyl moieties can be replaced by a wide range of others, as taught by the indicated publications, to give other DP-IV inhibitors for use in the present invention.

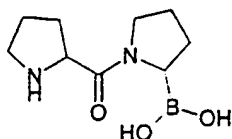
Amino-acyl pyrrolidides and thiazolidides (see DD 296 075 A5), e.g.



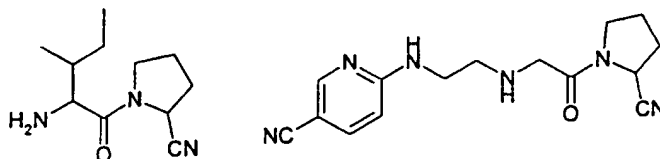
Amino-acyl pyrrolidine aldehydes (see DD 296 075 A5 and WO95/15309), e.g.



Amino-acyl pyrrolidine boronic acids (see WO91/16339 and WO93/08259), e.g.



Amino-acyl pyrrolidine nitriles (see WO95/15309 and WO98/19998), e.g.



In a preferred embodiment of the invention, the inhibitor of DP-IV is an amino-acylpyrrolidine nitrile. Particularly preferred are those amino-acyl pyrrolidine nitriles disclosed in WO95/15309 and WO98/19998.

The compositions according to the present invention may be formulated for administration to human subjects by any of the known routes, including oral administration, transmucosal administration (such as buccal, sublingual, intranasal, vaginal and rectal administration), transdermal administration or injection (including intravenous, intramuscular and subcutaneous injection). A preferred route of administration is oral administration. In this case the composition is suitably formulated as a tablet or capsule.

In a second aspect, the present invention comprises a new use for compounds that are known to be inhibitors of DP-IV, which is as therapeutic agents for the treatment of sub-normal development and dwarfism or for the promotion of tissue regeneration.



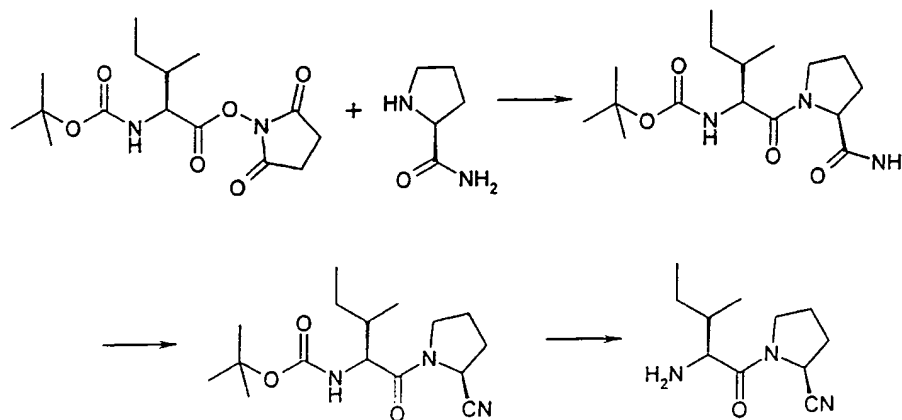
In a third aspect, the present invention comprises an improved method for the treatment of sub-normal development and dwarfism, particularly that due to growth hormone deficiency, and for the promotion of tissue regeneration, wherein the patient is administered a pharmaceutical composition comprising a therapeutically effective amount of inhibitor of DP-IV. The treatment may involve the use of said composition alone or in conjunction with other agents such as have been described heretofore. The administration may be as a single dose or as divided doses taken at intervals of, for example, 2 - 6 hours. In a particularly preferred dosing schedule, the composition is given once per day in the evening prior to sleep. The course of treatment might last for a period of a few days or weeks when used to promote tissue regeneration, or for a longer period when used to treat sub-normal development. The responsible physician will determine when a suitable clinical endpoint has been reached, as well as the details of the dosing regimen.

## Examples

### Example 1. Preparation of Inhibitors

The inhibitors of DP-IV can be prepared following the methods outlined on the literature. The synthesis of amino-acyl pyrrolidine nitriles is described in WO95/15309 and WO98/19998. The following method is illustrative of these methods.

#### Example 1A - Synthesis of (2S)-N-isoleucylpyrrolidine-2-carbonitrile.



(a) *tert*-Butyloxycarbonyl-isoleucylprolinamide

To a stirred suspension of prolinamide hydrochloride (225mg, 1.50mmol) in dry dichloromethane (15mL) was added diisopropylethylamine to give a clear basic (pH 9) solution. *N*-(*tert*-Butyloxycarbonyl-isoleucyloxy)succinimide (328mg, 1.0mmol) was added in one portion and the mixture was stirred at room temperature for 16 hours under a nitrogen atmosphere. The solvent was evaporated *in vacuo* and the residue was partitioned between ethyl acetate and 0.3*N* potassium hydrogensulphate solution. The organic layer was washed with saturated sodium hydrogencarbonate solution, water and brine, dried over sodium sulphate, and concentrated *in vacuo*. The residue was purified by filtration through a short plug of silica gel, eluting with hexane/ethyl acetate (10:90) then ethyl acetate. Concentration of the product-containing eluate gave the title compound as a colourless foaming glass; 301mg (92%).

<sup>1</sup>H NMR (CDCl<sub>3</sub>): δ 6.90 (1H, br. s); 5.51 (1H, br. s); 5.18 (1H, d, J=9.6Hz); 4.62 (1H, dd, J=2.6 & 7.0Hz); 4.29 (1H, dd, J=8.4 & 9.2Hz); 3.79-3.58 (2H, m); 2.36 (1H, m); 2.09-1.57 (5H, m); 1.43 (9H, s); 1.17 (1H, m); 0.95 (3H, d, J=6.6Hz); 0.90 (3H, t, J=7.3Hz) ppm.

(b) (2*S*)-*N*-(*tert*-Butyloxycarbonyl-isoleucyl)pyrrolidine-2-carbonitrile

To a stirred solution of the amide of part (a) (203mg, 0.62mmol) in dry pyridine (10mL) under a nitrogen atmosphere was added imidazole (84mg, 1.24mmol). The mixture was cooled to -35°C and then phosphorus oxychloride (0.25mL, 2.48mmol) was added dropwise. The mixture was stirred for 1 hour, during which time the temperature was allowed to rise to -20°C, and the solvent was evaporated *in vacuo*. The residue was purified by chromatography on silica gel to give the title compound as a colourless oil; yield 180mg (94%).

<sup>1</sup>H NMR (CDCl<sub>3</sub>): δ 5.14 (1H, d, J=9.2Hz); 4.80 (1H, dd, J=2.6 & 7.1Hz); 4.22 (1H, dd, J=7.9 & 9.1Hz); 3.81 (1H, m); 3.71 (1H, m); 2.30-2.12 (4H, m); 1.75 (1H, m); 1.60 (1H, m); 1.42 (9H, s); 1.19 (1H, m); 0.97 (3H, d, J=6.9Hz); 0.91 (3H, t, J=7.3Hz) ppm.

<sup>13</sup>C NMR (CDCl<sub>3</sub>): δ 171.7; 155.6; 118.0; 79.6; 56.0; 46.5; 46.0; 37.8; 29.6; 28.1; 25.0; 24.2; 15.2; 10.9 ppm.

(c) (2S)-N-(Isoleucyl)-pyrrolidine-2-carbonitrile trifluoroacetate

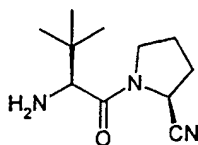
The nitrile of part (b) was dissolved in trifluoroacetic acid and the solution was stirred at room temperature for 1 hour. The solvent was evaporated *in vacuo* and the residue was dissolved in water. The solution was lyophilised to give the title compound as a white fluffy solid; yield 60mg.

FAB Mass Spec.: Calculated m/e 209.3; Found 210.2 (M+H)<sup>+</sup>

<sup>1</sup>H NMR (D<sub>2</sub>O): δ 4.3 (1H, m); 3.64 (1H, d, J=5.6Hz); 3.16 (2H, m); 1.86-1.48 (5H, m); 0.98 (1H, m); 0.68 (1H, m); 0.51 (3H, d, J=6.9Hz); 0.38 (3H, t, J=7.3Hz) ppm.

<sup>13</sup>C NMR (D<sub>2</sub>O): δ 169.7; 119.7; 57.3; 48.6; 48.1; 36.9; 30.2; 25.8; 24.5; 15.4; 11.5 ppm.

Example 1B - Synthesis of (2S)-N-((2S)-2'-amino-3',3'-dimethylbutanoyl)pyrrolidine-2-carbonitrile.



This was prepared following the method of Example 1A by replacing the isoleucine derivative with the corresponding *tert*-butylglycine derivative.

<sup>1</sup>H NMR (CD<sub>3</sub>OD): δ 4.86-4.81(1H, m); 4.04(1H, s); 3.77-3.71(2H, m); 3.34(2H, s); 2.34-2.08(4H, m); 1.14(9H, s) ppm.

<sup>13</sup>C NMR (CD<sub>3</sub>OD): δ 167.40, 117.99, 58.78, 46.53, 34.21, 29.54, 25.22, 25.03 ppm.

## Example 2. Effect on circulating Growth Hormone

### *Animals*

Experiments were conducted in male Sprague-Dawley rats (200-220g) obtained from Iffa Credo (L'Arbresle, France), fed with standard laboratory chow *ad libitum* and kept on a 12 h light-dark in a temperature- and humidity-controlled room.

### *Surgery*

Rats were weighed and anesthetized with Narcorene. The ventral side of the throat was shaved and an incision was made down the center of the throat. The right jugular vein was exposed and cannulated using a polythene tubing (OD 1.0mm) connected to a medical grade silicone tubing (OD 0.94mm) (silicone side in the jugular vein). The tubing was secured and the polythene side was externalized through an incision made on the dorsal side of the neck. The catheter was rinsed with 300µL of a Ringer solution containing 0.1% heparin. Rats were allowed to recover for at least 24 hours in individual cages with food and water available *ad libitum*.

### *Experimental procedure*

The compound of Example 1B (3mg/kg, i.v.) or its vehicle was injected 15 minutes before an injection of GHRH (0.1µg/kg, i.v.). Blood samples (200-250µL) were removed immediately before inhibitor and GHRH injections, and then 5, 10, 15, 20, 30 and 60min post-GHRH. Each blood sample was replaced with an equivalent volume of a Ringer solution containing 0.1% heparin. Plasma was extracted and stored at -20°C until determinations of GH by RIA.

### *Results*

The compound of Example 1B potentiated the GH surge induced by the administration of GHRH (Table 1). Integrated GH responses were  $720.2 \pm 217.0$  ng/ml.60 min and  $5072.8 \pm 837.3$  ng/ml.60 min in the vehicle- and inhibitor-treated group respectively ( $p < 0.05$ ).

**Table 1:** Effects of the DP-IV inhibitor of Example 1B on the GH surge induced by an intravenous injection of GHRH. Results are mean GH ng/ml of 6 rats  $\pm$  s.e.m.

	Time (min)								AUC
	-15	0	5	10	15	20	30	60	
GHRH	42.1 $\pm 12.5$	40.1 $\pm 8.0$	155.7 $\pm 25.1$	91.0 $\pm 15.6$	61.0 $\pm 12.9$	53.6 $\pm 12.5$	34.1 $\pm 8.2$	26.7 $\pm 3.0$	720.2 $\pm 217.0$
GHRH + Inhibitor	50.5 $\pm 16.4$	33.4 $\pm 6.9$	483.0 $\pm 91.0$	309.5 $\pm 55.40$	164.3 $\pm 28.3$	103.1 $\pm 18.1$	52.1 $\pm 7.7$	26.3 $\pm 3.3$	5072.8 $\pm 837.3$

The results obtained indicate that inhibitors of DP-IV are able to increase circulating levels of growth hormone and hence are useful in the treatment of sub-normal development due to growth hormone deficiency and other conditions where the promotion of tissue growth is important.

### Example 3. Pharmaceutical formulation

#### 3A – 50mg Tablet

Tablets containing the equivalent of 50mg of the compound of Example 1A as the active agent are prepared from the following:

Compound of Example 1A (as trifluoroacetate salt)	154.5g
Corn starch	53.5g
Hydroxypropylcellulose	13.5g
Carboxymethylcellulose calcium	11.0g
Magnesium stearate	2.0g
Lactose	165.5g
<i>Total</i>	<i>400.0g</i>

The materials are blended and then pressed to give 2000 tablets of 200mg, each containing the equivalent of 50mg of the free base of the compound of Example 1A.

3B – 100mg Vaginal suppository

Suppositories suitable for vaginal administration and containing the equivalent of 100mg of the compound of Example 1A as the active agent are prepared from the following:

Compound of Example 1A (as trifluoroacetate salt)	154.5g
Corn starch	210.0g
Colloidal silica	2.5g
Povidone 30	49.0g
Magnesium stearate	23.0g
Adipic acid	57.0g
Sodium bicarbonate	43.0g
Sodium lauryl sulphate	5.0g
Lactose	456.0g
<hr/> Total	<hr/> 1000.0g

The materials are blended and then pressed to give 1000 suppositories of 1g, each containing the equivalent of 100mg of the free base of the compound of Example 1A.

The foregoing Examples are illustrative of the invention as disclosed herein, but are not intended to be limiting. Such extensions as would be considered equivalent by one skilled in the art are included within the scope of the invention and the Claims that further define that scope.

One or more DP-IV inhibitors may be used as the sole component active for the specified purposes of the composition and method of the invention.

**Claims**

1. A pharmaceutical composition for the treatment of sub-normal growth or dwarfism, which composition is characterized in that it comprises a therapeutically effective amount of inhibitor of dipeptidyl peptidase IV.
2. A composition according to Claim 1, wherein the sub-normal growth or dwarfism is due to growth hormone deficiency.
3. A pharmaceutical composition for the promotion of tissue regeneration, which composition is characterized in that it comprises a therapeutically effective amount of inhibitor of dipeptidyl peptidase IV.
4. A composition according to any preceding claim, wherein the inhibitor of dipeptidyl peptidase IV comprises amino-acyl pyrrolidine nitrile.
5. A composition according to any preceding claim, wherein the composition is formulated for oral administration.
6. The composition according to Claim 5 formulated as a tablet or capsule.
7. The use of inhibitor of dipeptidyl peptidase IV in the preparation of a therapeutic composition for the treatment of sub-normal development or dwarfism or for the promotion of tissue regeneration.
8. A method for the treatment of sub-normal development or dwarfism, wherein the patient is administered a pharmaceutical composition comprising inhibitor of dipeptidyl peptidase IV.
9. A method according to Claim 8, wherein the composition is administered once per day in the evening.
10. A method of tissue regeneration wherein the patient is administered a pharmaceutical composition comprising inhibitor of dipeptidyl peptidase IV.

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DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID,  
IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT,  
LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL,  
PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ,  
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- (84) Designated States (regional): ARIPO patent (GH, GM,  
KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent  
(AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent  
(AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU,  
MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM,  
GA, GN, GW, ML, MR, NE, SN, TD, TG).
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- For two-letter codes and other abbreviations, refer to the "Guid-  
ance Notes on Codes and Abbreviations" appearing at the begin-  
ning of each regular issue of the PCT Gazette.



WO 00/56297 A3

(54) Title: PHARMACEUTICAL COMPOSITIONS COMPRISING DIPEPTIDYL PEPTIDASE IV INHIBITORS FOR THE PRO-  
MOTION OF GROWTH

(57) Abstract: Inhibitors of dipeptidyl peptidase IV and pharmaceutical compositions comprising these inhibitors are useful in the  
treatment of short stature due to Growth Hormone deficiency and for promoting GH-dependent tissue growth or regrowth.



## INTERNATIONAL SEARCH REPORT

International Application No.

PCT/IB 00/00393

A. CLASSIFICATION OF SUBJECT MATTER  
IPC 7 A61K31/40 A61P5/00

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 19998 A (CIBA GEIGY AG ;VILLHAUER EDWIN BERNARD (US)) 14 May 1998 (1998-05-14) cited in the application page 18, line 19-21 claims; examples ---	1-7, 10
X	WO 93 08259 A (NEW ENGLAND MEDICAL CENTER INC ;UNIV TUFTS (US)) 29 April 1993 (1993-04-29) cited in the application page 21 --- -/--	1-6

☒ Further documents are listed in the continuation of box C.☒ Patent family members are listed in annex.

## \* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- "&" document member of the same patent family

Date of the actual completion of the international search

27 October 2000

Date of mailing of the international search report

08/11/2000

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Authorized officer

Veronese, A

## INTERNATIONAL SEARCH REPORT

Internal Application No

PCT/IB 00/00393

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	REYNOLDS: "MARTINDALE The Extra Pharmacopoeia", ROYAL PHARMACEUTICAL SOCIETY, LONDON XP002151318 page 1271, column 3 -page 1272, column 2 page 1283, column 1 -page 1284, column 3 page 1293, column 2-3 ---	1,2,4,7, 8
Y	ASHWORTH, DOREEN M. ET AL: "2-Cyanopyrrolidides as potent, stable inhibitors of dipeptidyl peptidase IV" BIOORG. MED. CHEM. LETT. (1996), 6(10), 1163-1166, XP000953254 the whole document ---	1,2,4,7, 8
Y	BONGERS J ET AL: "KINETICS OF DIPEPTIDYL PEPTIDASE IV PROTEOLYSIS OF GROWTH HORMONE -RELEASING FACTOR AND ANALOGS." BIOCHIM BIOPHYS ACTA, (1992) 1122 (2), 147-153., XP000953283 the whole document ---	1,2,4,7, 8
P,Y	MENTLEIN, ROLF (1): "Dipeptidyl-peptidase IV (CD26)-role in the inactivation of regulatory peptides." REGULATORY PEPTIDES, (NOV. 30, 1999) VOL. 85, NO. 1, PP. 9-24., XP000953272 See figure 4: rate of cleavage of GRH by DPPIV page 16, column 1-2 ---	1,2,4,7, 8
A	CAMPBELL, ROBERT M. (1) ET AL: "Enhanced stability and potency of novel growth hormone-releasing factor (GRF) analogues derived from rodent and human GRF sequences." PEPTIDES (TARRYTOWN), (1994) VOL. 15, NO. 3, PP. 489-495., XP000953247 page 494, column 2, line 18-20 ---	1-10
A	PRAGER M.D. ET AL: "Dipeptidyl peptidase IV and aminopeptidase in burn wound exudates: Implications for wound healing." JOURNAL OF TRAUMA, (1994) 36/5 (629-633). XP000953245 the whole document -----	3,10

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

## Continuation of Box I.2

Present claims 1-3,5-10 relate to a rather large number of possible compounds, which are defined by reference to a pharmacological mechanism of action: "inhibitor of dipeptidyl peptidase IV". The definition of compounds by reference to a pharmacological mechanism in the present context is considered to lead to a lack of clarity within the meaning of Art. 6 PCT. It is impossible to fully compare the parameters the applicant has chosen to employ with what is set out in the prior art. Also the definition "amino-acyl pyrrolidine nitrile" in claim 4 relates to a rather elevated number of possible compounds. Support and/or disclosure within the meaning of Articles 5,6 PCT is to be found however for only a very small proportion of the compounds claimed.

Also the expression "tissue regeneration" in claim 3, 7, 10 comprises numerous diseases. The claims covers all such diseases, whereas the application provides support and/or disclosure within the meaning of Articles 5, 6 PCT for only a very small proportion of the diseases claimed.

Consequently, the search has been carried out for those parts of the claims which appear to be clear, concise, supported and disclosed, namely for the amino - acyl pyrrolidine nitrile derivatives shown in the examples of present application and disclosed in the cited applications W09515309 and W09819998, in respect to the treatment of dwarfism and the expression "tissue regeneration" as such, (no other diseases have been searched), with due regard to the general idea underlying the application.

All claims 1 to 10 have been searched incompletely.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/IB 00/00393

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9819998 A	14-05-1998	AU 5318498 A	29-05-1998
		BR 9714130 A	29-02-2000
		CN 1236361 A	24-11-1999
		CZ 9901615 A	11-08-1999
		EP 0937040 A	25-08-1999
		NO 992028 A	28-04-1999
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